The association between cardiovascular risk factors and C-reactive protein differs between gender

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Abstract

Background. The aim of the present study was to study, first, whether C-reactive protein (CRP) levels differ between genders. Second, to explore which cardiovascular (CV) risk factors, including urinary albumin excretion (UAE), are associated with an elevated CRP level and third, whether these associations differ between men and women.

Methods and Results. The association between CV risk factors and CRP was studied in 3,969 men and 3,757 women. Risk factors studied were higher age, hypertension, hypercholesterolemia, measures of obesity, smoking, positive family history for CV disease, UAE, and the use of oral contraceptives (OC) and hormone replacement therapy (HRT) in women. CRP and UAE were dichotomised into the lowest four quintiles and the highest quintile stratified for gender. In a multivariate analysis, the use of OC (OR 3.6 [2.7-4.6]), BMI >30 kg/m2 (2.6 [2.0-3.5]) and abdominal obesity (2.7 [2.1-3.5]) contributed most strongly to an elevated CRP in women. In men, smoking (2.3 [1.9-2.8]) was, besides age (1.9 [1.5-2.3]), the most predominant factor contributing to an elevated CRP. These associations did not change when including UAE in the model.

Conclusions. We conclude that the risk factors contributing to an elevated CRP differ between men and women. This is probably due to hormonal and obesity-related factors. CRP and UAE may reflect different pathways leading to CV disease.

Introduction

Various risk factors are known to contribute to an increased risk of cardiovascular (CV) morbidity and mortality, such as for example hypertension, diabetes, obesity, hyperlipidemia and smoking. It is however not clear yet, which patient will suffer from these risk factors and which will not. At present therefore, much attention focuses on the determination of risk markers, which can be considered as surrogate end points of early atherosclerotic damage. To that purpose, C-reactive protein (CRP),[1-4] fibrinogen,[5] urinary albumin excretion (UAE),[3,4,6] and carotid intima thickness[7] are being used.

Elevated CRP levels[8-13] as well as microalbuminuria[14-18] indeed have been found in association with older age, hypertension, diabetes, obesity, hyperlipidemia, the insulin resistance syndrome, and smoking, as well as oral contraceptive use and hormone replacement therapy. An elevated CRP level is suggested to reflect vascular low-grade inflammation characteristic of atherosclerosis,[2] while microalbuminuria seems more to be a marker of generalised vascular or endothelial damage.[19]

CV morbidity and mortality are more prevalent in men than in women.[20] The exact mechanism of

this difference between the genders is not known. It may be related to a higher prevalence of known CV risk factors in men, but also to a difference in susceptibility for these risk factors. Interestingly however, CRP has been found higher in women than in men.[13] The aim of the present study was therefore, first to study whether CRP indeed differs between the genders, second, to explore which CV risk factors, including UAE, are associated with an elevated CRP level and third, whether these associations differ between men and women.

Methods

Study Population

This study is part of the ongoing 'PREVEND' study (Prevention of REnal and Vascular ENd stage Disease), in the city of Groningen, the Netherlands. All inhabitants, aged 28-75 years (n=85,421), were asked to send in a morning urine sample and to fill out a short questionnaire on demographics and CV history. 40,856 subjects (47.8%) responded. Our actual study cohort was derived from this cohort and based on all subjects with a urinary albumin concentration of ≥10mg/L (n=7,768) in their morning urine together with a randomly selected control group with a urinary albumin concentration of <10 mg/L (n=3,395). Details of this protocol have been described elsewhere.[18,21] In total 11,163 subjects were invited to the outpatient clinic, of which 8,592 subjects completed the screening program. Subjects using insulin or pregnant women were excluded from participation in this screening program. The study was approved by the medical ethics committee and conducted in accordance with the guidelines of the declaration of Helsinki. All participants gave written informed consent.

Study Design

The screening program in the outpatient clinic consisted of two visits. At the first visit, participants completed a self-administered questionnaire regarding demographics, CV and renal history, and the use of blood pressure and lipid lowering drugs. After removal of shoes and heavy clothing, weight was measured to the nearest 0.5 kg with a Seca balance scale (Seca Vogel & Halke GmbH & Co., Hamburg, Germany). Height was measured to the nearest 0.5 cm. Waist circumference was measured between the superior iliac crest and the lowest rib to the nearest 0.5 cm. At both visits, blood pressure was measured in supine position, every minute, for 10 and 8 minutes respectively, with an automatic Dinamap XL Model 9300 series monitor (Johnson-Johnson Medical INC, Tampa, Florida, USA). Subjects were asked to collect 24-hour urine on two consecutive days in the last

week before the second visit. The subjects were given oral and written instructions on how to collect 24-hour urine and they were instructed to postpone urine collection in case of fever, urinary tract infection, and menstruation. Furthermore, they were asked to refrain as far as possible from heavy exercise during the collection period. The subjects were asked to store the urine cold (4°C) for a maximum of four days prior to the second visit. Measurements of urinary volume and albumin concentration were performed on each collection. At the second visit, blood was drawn after an overnight fast, for determination of plasma glucose, serum C-reactive protein and cholesterol. Community pharmacy data on the use of contraceptive drugs and hormone replacement therapy were collected for each subject as described previously.[22]

Calculations

Systolic and diastolic blood pressure was calculated as the mean of the last two measurements of the two visits. Body mass index (BMI) was calculated as the ratio between weight and the square of height (kg/m^2). Urinary albumin excretion is given as the mean of the two 24-hour urine excretions (mg/24hr).

Laboratory Methods

High sensitive C-reactive protein was measured by nephelometry with a threshold of 0.18 mg/L and intra-and interassay coefficients of variation of <4.4 and <5.7%, respectively (BNIIN, Dade Behring, Marburg, Germany). Urinary albumin concentration was determined by nephelometry with a threshold of 2.3 mg/L and intra- and inter-assay coefficients of variation of <2.2% and <2.6%, respectively (Dade Behring, Marburg, Germany). Plasma glucose and serum cholesterol were determined by Kodak Ektachem dry chemistry (Eastman Kodak, Rochester, New York, USA). Urinary leukocyte and erythrocyte measurements were done by Nephur-test+leuco sticks (Boehringer Mannheim, Mannheim, Germany).

Data handling and definitions

In the present analysis we excluded 451 subjects because of erythrocyturia or leukocyturia (erythrocytes >50 cells/mm^3 or leukocytes >75 cells/mm^3, or leukocytes = 75 cells/mm^3 and erythrocytes >5 cells/mm^3). In 415 subjects, CRP could not be measured. All together, 7,726 subjects were eligible for this analysis.

Risk factors studied were higher age, hypertension, anti-hypertensive therapy, measures of

obesity, hypercholesterolemia, statin use, myocardial infarction, diabetes, positive family history for CV disease, smoking and UAE, as well as the use of hormone replacement therapy (HRT) and oral contraceptive drug use in women. Higher age was defined as an age of >50 years (median 49 years). Hypertension was defined as a systolic blood pressure ≥140 mmHg or a diastolic blood pressure of ≥90 mmHg. Obesity was defined as a body mass index of >30 kg/m^2.[23] Increased central fat distribution (abdominal obesity) was defined as a waist circumference of ≥102 cm in men and ≥88 cm in women.[23] Hypercholesterolemia was defined as a plasma cholesterol of ≥6.5 mmol/L. A history of myocardial infarction was considered present if the subject reported having been hospitalised for at least three days due to that condition. Diabetes was defined as fasting plasma glucose levels ≥7.0 mmol/L, or non-fasting plasma glucose levels ≥11.1 mmol/L or the use of oral anti-diabetic drugs.[24] The history for CV disease was found positive when a first-grade relative had experienced a CV event before 55 years of age. Smokers were defined as currently smoking or quit smoking within the last year.

Oral contraceptives were defined as preparations containing ethinylestradiol and a progestin. Hormone replacement therapy was defined as oral preparations containing conjugated estrogens or estradiol valerate, or transdermal preparations containing estradiol. Vaginal preparations containing estriol or dienestriol were not considered hormone replacement therapy.

Statistical analysis

Continuous data are reported as mean with standard deviation. In case of a skewed distribution, the median and its interquartile range are presented. CRP and UAE were categorised into quintiles based on cut-off points stratified for gender. CV risk factors were compared over various ranges of CRP separately for gender and subsequently between men and women. The untransformed distributions of CRP and UAE were skewed; therefore, natural logarithmic transformation was applied before further analyses. Significance levels were determined by one way analysis of variance in case of Gaussian distributions and by Mann-Whitney rank sum test in case of non-Gaussian distributions. Chi square analysis or Fisher's exact test were carried out when appropriate. In addition, interactions between gender, the level of CRP and the studied risk factor were tested in the entire population with a two way ANOVA or Breslow-Day and Tarone's statistics for testing the homogeneity of the common odds ratio when appropriate. The *P* value for testing inequality among groups was computed. The level of significance was determined at alpha < 0.05.

Logistic regression analysis was used in men and women separately. CRP was dichotomised into the lowest four quintiles against the highest quintile, adjusted for age and sex, adjusted for all risk factors without UAE, and adjusted for all risk factors including UAE to find the strongest

determinants of an increased CRP level. Finally, a multivariate logistic regression analysis, including variables for interaction between the genders and other CV risk factors, was performed. SPSS for Windows 10.0 was used for all statistical analyses.

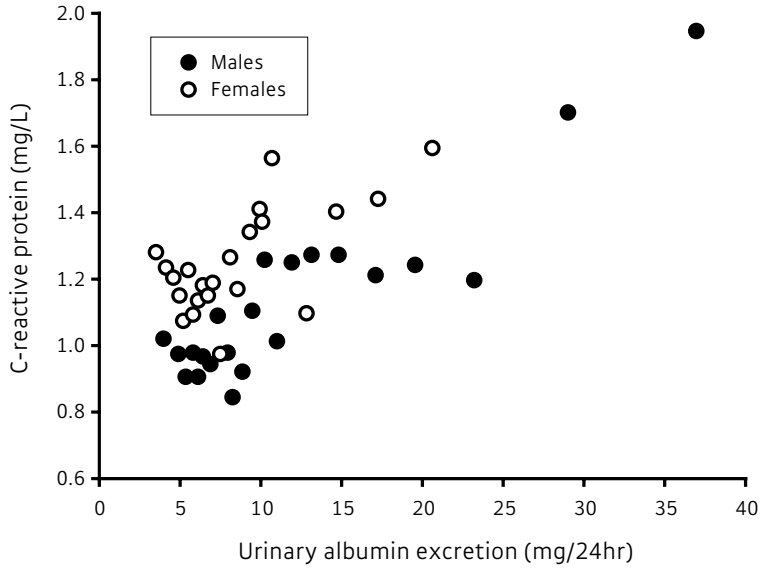
Results

The sample comprised 3,969 men and 3,757 women. The characteristics of these subjects stratified for gender are summarised in Table 1. Among the continuous variables, age, blood pressure, BMI, waist circumference, plasma cholesterol, glucose and UAE were higher in men than in women, whereas CRP was slightly higher in women. Risk factors were generally more prevalent in men than in women, except for the prevalence of an elevated CRP, and obesity, in particular abdominal obesity, which were higher in women. The prevalence of smoking and a positive family history for CV disease did not differ between men and women.

Figure 1 shows the association between UAE and CRP in men and women separately. UAE is divided into 20-tiles and median CRP values for the corresponding percentile are presented. A higher UAE was associated with a higher CRP in both genders. For a given UAE, CRP levels were higher in women than in men. Gender-specific characteristics classified according to CRP quintiles are presented in table 2. The percentage of subjects with a certain CV risk factor varied widely across the various CRP quintiles, with increasing prevalences of all risk factors in both genders in the higher CRP quintiles (*P* values <0.05), except for HRT in women (*P*=0.131). The association between a number of risk factors with an elevated CRP differed between gender: older age increased more in men (from 21.6% to 65.1%) than in women (from 21.8% to 45.2%; *P*<0.001), actual smokers increased more in men (from 24.1% to 51.5%) than in women (from 33.5 to 39.9%; *P*<0.001). Also, the percentage of subjects with elevated UAE increased more in men than in women (from 8.3 to 34.6% and from 12.9 to 29.6%; *P*=0.004) On the other hand, the percentage of obesity and abdominal obesity increased more in women (from 9.5 to 56.2% and from 2.1% to 36.4%) than in men (from 5.0 to 37.2% and from 3.1% to 23.8%; both *P*<0.001).

Table 3 presents the results of the gender-specific multivariate analysis examining the association of the CV risk factors of interest with an elevated CRP level. It first shows that in men most CV risk factors were independently associated with an increased risk of having an elevated CRP level (if confidence interval is OR ≥1). Statin use was associated with a lower risk of having an elevated CRP. In women, age, antihypertensive medication, high cholesterol levels, statin use, history of myocardial infarction, a positive family history for CV disease, HRT use and an elevated UAE were not associated with an increased CRP level. It second shows that in women, the use of

Figure 1. Association between urinary albumin excretion and C-reactive protein levels in males and females.



oral contraceptives (OR 3.6 [2.7-4.6]), obesity (2.6 [2.0-3.5]) and abdominal obesity (2.7 [2.1-3.5]) were the risk factors that most strongly contributed to an increased risk for an elevated CRP, also expressed by the Wald statistic. In contrast, smoking (2.3 [1.9-2.8]) and older age (1.9 [1.5-2.3]) were the most important risk factors associated with a CRP in the upper quintile in men. The table finally shows that increased UAE was also associated with an increased CRP, but that adding UAE into the model did not significantly influence the association between any of the risk factors and CRP.

Table 4 shows the results of the multivariate logistic regression analysis for the complete data set, including variables for interaction between gender and the CV risk factors. We observed interactions between gender and age, gender and smoking, and gender and measures of obesity. These interactions are graphically depicted in figure 2. In this figure, males without the risk factor of interest are taken as the reference category (OR=1, front bar). It shows that an elderly male was ~2-fold as likely to have an elevated CRP compared to women (2a). Smoking males were ~2-fold as likely to have an elevated CRP level compared to non-smoking women (2b). Compared to a non-obese male, peripheral and abdominal obese females were ~2.5-fold as likely to have an elevated CRP level (2c, 2d). To explore effect modification by post-menopausal status, we have conducted a stratified analysis controlling for age. The calculated adjusted estimates were roughly the same for both men and women.

Table 1. Characteristics of the study cohort, divided for men and women.

Risk factors	Male (n=3969)	Female (n=3757)	Total (n=7726)	*P* value†
Continuous variables				
Age, yr	50.2 ± 12.9	47.8 ± 12.2	48.0 ±12.6	<0.001
Systolic blood pressure, mmHg	133.6 ± 18.5	124.0 ± 20.8	129.0 ± 20.2	<0.001
Diastolic blood pressure, mmHg	76.8 ± 9.5	71.0 ± 9.1	74.0 ± 9.8	<0.001
Body mass index, kg/m^2	26.3 ± 3.7	25.8 ± 4.7	26.0 ± 4.2	<0.001
Waist circumference, cm	93.8 ± 11.1	82.8 ± 12.5	88.5 ± 13.0	<0.001
Cholesterol, mmol/L	5.7 ± 1.1	5.6 ± 1.1	5.6 ± 1.1	0.001
Glucose, mmol/L	5.0 ± 1.3	4.7 ± 1.1	4.9 ± 1.2	<0.001
CRP, mg/L	1.2 (0.6-2.7)	1.3 (0.5-3.2)	1.3 (0.2-2.9)	<0.008
UAE, mg/24hr	11.5 (6.8-22)	8.1 (5.7-14)	9.2 (6.2-17.2)	<0.001
Dichotomous variables				
Age >50 years, %	46.1	38.4	42.4	<0.001
SBP ≥140 and/or DBP ≥90, %	32.5	21.1	27.0	<0.001
Antihypertensive medication, %	12.8	11.0	11.9	0.016
Obesity, %	14.3	16.1	15.1	0.030
Abdominal obesity, %	23.0	31.5	27.1	<0.001
Cholesterol ≥6.5 mmol/L	25.3	23.7	24.5	0.100
Statin use, %	5.8	3.8	4.8	0.001
Myocardial infarction, %	8.5	4.0	6.4	<0.001
Diabetes mellitus, %	4.4	3.1	3.8	0.003
Family history, %	22.9	22.0	22.5	0.402
Smoking, %	37.7	37.3	37.5	0.724
Oral contraceptives, %	0	19.2	9.7	-
Hormone replacement therapy, %	0	3.5	1.8	-
Highest CRP quintile, %	18.3	21.7	20.0	<0.001
Highest UAE quintile, %	25.4	14.3	20.0	<0.001

Mean (standard deviation) and median values (25th-75th percentile) are presented in case of normal and skewed distributions, respectively. CRP = C-reactive protein, UAE = urinary albumin excretion, SBP = systolic blood pressure, DBP = diastolic blood pressure. † *P* value indicates whether means, medians or prevalence of a certain risk factor differs between men and women.

Table 2 (males). Characteristics of the study cohort, divided by C-reactive protein, for men and women separately.

Risk factors	CRP level (mg/L)						
	< 0.46	0.47-0.91	0.92-1.63	1.64-3.30	> 3.31	*P* value†	*P* value‡
Male							
N	783	801	795	797	793		
Age >50 y, %	21.6	36.2	49.7	57.6	65.1	<0.001	<0.001
SBP ≥140 and/or DBP ≥90, %	15.3	26.8	34.6	40.2	45.5	<0.001	0.325
Antihypertensive medication, %	4.7	6.8	12.8	17.8	21.9	<0.001	0.216
Obesity, %	3.1	9.9	13.1	21.4	23.8	<0.001	<0.001
Abdominal obesity, %	5.0	16.1	24.2	32.2	37.2	<0.001	0.002
Cholesterol ≥6.5 mmol/L	13.8	21.7	28.7	28.8	33.3	<0.001	0.449
Statin use, %	3.4	5.0	6.4	6.5	7.4	0.022	0.926
Myocardial infarction, %	5.3	5.2	6.7	11.0	14.6	<0.001	0.209
Diabetes mellitus, %	1.2	1.8	3.7	4.9	7.5	<0.001	0.740
Family history, %	19.8	20.1	22.1	25.7	27.1	0.003	0.999
Smoking, %	24.1	32.5	36.9	43.1	51.5	<0.001	<0.001
Highest UAE quintile, %	8.3	13.5	18.5	25.1	34.6	<0.001	0.004

† *P* value indicates significance level of the association between a risk factor and CRP (one way ANOVA test for continuous variables or chi square test in case of dichotomous variables). ‡ *P* value indicates the significance level of the interaction between gender, risk factor and the level of CRP (Breslow-Day and Tarone's statistics).

Discussion

We showed that the level of CRP in blood is higher in women than in men. Most of the CV risk factors were associated with an increased CRP level, but clear differences exist between the genders. In women, the use of oral contraceptives and measures of obesity had the highest impact, while in men, besides age, smoking, was the most important determinant of a higher CRP. We moreover showed that CRP levels increased with a higher UAE, but that the presence of an increased albumin excretion did not change the association between CV risk factors and CRP. We hypothesize that the association between CV risk factors and CRP does not include a mechanistic pathway in which UAE is involved.

Our findings that CRP levels were higher in women than in men is consistent with other data in the literature.[11,13] This is of interest, since CV risk factors were generally more prevalent in men than in women. In line with our data, a recent report among US women not taking HRT

Table 2 (females). Characteristics of the study cohort, divided by C-reactive protein, for men and women separately.

Risk factors	CRP level (mg/L)					
	<0.44	0.44-0.92	0.93-1.81	1.82-3.92	>3.92	*P* value†
Female						
N	744	753	754	755	750	
Age >50 y, %	21.8	33.6	44.3	47.0	45.2	<0.001
SBP ≥140 and/or DBP ≥90, %	8.1	15.3	23.1	24.4	34.7	<0.001
Antihypertensive medication, %	2.7	7.0	13.2	14.0	18.0	<0.001
Obesity, %	2.0	5.4	15.2	21.2	36.3	<0.001
Abdominal obesity, %	9.5	17.7	32.0	41.8	56.2	<0.001
Cholesterol ≥6.5 mmol/L	14.5	20.6	25.5	29.5	28.2	<0.001
Statin use, %	2.4	2.6	4.6	4.5	4.7	0.044
Diabetes mellitus, %	0.5	1.2	2.0	3.8	8.0	<0.001
Myocardial infarction, %	3.8	2.7	3.3	5.0	5.4	0.015
Family history, %	19.1	19.2	21.2	25.1	25.5	0.007
Smoking, %	33.5	35.8	37.7	39.7	39.9	0.039
Oral contraceptives, %	11.4	12.9	18.0	23.5	29.7	<0.001
Hormone replacement therapy, %	2.3	3.1	3.6	4.8	3.7	0.131
Highest UAE quintile, %	12.9	15.1	19.8	22.5	29.6	<0.001

† *P* value indicates significance level of the association between a risk factor and CRP (one way ANOVA test for continuous variables or chi square test in case of dichotomous variables).

also showed that waist circumference, blood pressure, but not age and smoking status most prominently associated with an elevated CRP.[25] In a separate analysis, the same author earlier showed that, among US men aged >20 years, CRP levels did increase with advancing age and current smoking,[26] which is also in line with our report. However, interactions could not be performed in these studies.

Our finding that the use of oral contraceptives is the strongest determinant of a higher CRP is noteworthy. In one study by Doring *et al*, third generation oral contraceptive use was associated with an elevated CRP.[27] It suggests that the difference in hormonal situation between men and women partially explain the higher CRP in women. This finding is the more relevant, as the prevalence of CV risk factors was mostly lower in women than in men.

The association between higher age and CRP was stronger in men than in woman. Apparently,

Table 3 (males). Gender-specific logistic regression analysis relating cardiovascular risk factors to an increased C-reactive protein.

Risk factors	Crude	Age adjusted	Multivariate without UAE	Multivariate with UAE	Wald statistic
Male					
Age >50 yr	2.6 (2.2-3.1)		2.0 (1.6-2.5)	1.9 (1.5-2.3)	29.1
SBP ≥140 and/or DBP ≥90	2.0 (1.7-2.4)	1.5 (1.3-1.8)	1.3 (1.0-1.6)	1.2 (0.9-1.5)	2.2
Antihypertensive medication	2.4 (1.9-2.9)	1.7 (1.4-2.1)	1.3 (1.0-1.7)	1.3 (1.0-1.7)	2.7
Obesity	2.3 (1.9-2.8)	2.1 (1.7-2.6)	1.4 (1.0-1.9)	1.4 (1.0-1.9)	3.7
Abdominal obesity	2.5 (2.0-2.9)	2.0 (1.7-2.4)	1.5 (1.1-2.0)	1.4 (1.1-1.9)	6.3
Cholesterol ≥6.5 mmol/L	1.6 (1.4-1.9)	1.4 (1.2-1.7)	1.4 (1.1-1.8)	1.4 (1.1-1.8)	7.0
Statin use	1.4 (1.0-1.9)	1.1 (0.8-1.5)	0.7 (0.4-1.1)	0.7 (0.4-1.1)	3.0
Myocardial Infarction	2.3 (1.8-2.9)	1.7(1.3-2.2)	1.5 (1.1-2.2)	1.4 (1.0-2.0)	4.8
Diabetes	3.5 (2.6-4.8)	2.5 (1.8-3.5)	2.6 (1.7-3.8)	2.3 (1.5-3.4)	16.2
Family history	1.2 (1.0-1.5)	1.2 (1.0-1.5)	1.2 (0.9-1.5)	1.2 (0.9-1.5)	1.7
Smoking	2.0 (1.7-2.4)	2.3 (1.9-2.7)	2.4 (1.9-2.9)	2.3 (1.9-2.8)	63.7
Highest UAE quintile UAE	2.5 (2.0-2.9)	2.1 (1.8-2.6)		1.8 (1.4-2.2)	21.8

Crude, age adjusted and multivariate gender-specific logistic regression models with and without elevated UAE in the model relating CV risk factors to the risk of having an elevated CRP level for that specific risk factor. The Wald statistic indicates the strength of the association.

in men, CRP levels rise with age. In women, some confounding factors may play a role. It may well be that menopausal status is major player: CRP is likely to be increased in pre-menopausal women due to their endogenous estrogen status (including OC use), while in postmenopausal women the effect of risk factors and ageing will likely yield increments of CRP. Indeed, after controlling for CV risk factors and OC use, the risk of having an elevated CRP level was found equal between younger and elderly women (figure 2a). In another study by Sites *et al*, CRP did not differ by menopausal status either.[28] To partly overcome the possibility of confounding by menopausal status, we stratified our analyses for age. This method however, did not change the associations as well as the observed interactions.

The finding that obesity contributes most importantly to a higher CRP in women is supported by data from three other studies that showed a stronger association between BMI and CRP in women when compared with men.[13,29,30] Obesity, and more specifically abdominal obesity, is an important hallmark of the insulin resistance syndrome. Interestingly, obesity as measured by

Table 3 (females). Gender-specific logistic regression analysis relating cardiovascular risk factors to an increased C-reactive protein.

Risk factors	Crude	Age adjusted	Multivariate without UAE	Multivariate with UAE	Wald statistic
Female					
Age >50 yr	1.4 (1.2-1.7)		1.0 (0.8-1.4)	1.0 (0.8-1.4)	0.10
SBP ≥140 and/or DBP ≥90	2.5 (2.0-2.9)	2.4 (1.9-2.9)	1.7 (1.3-2.2)	1.7 (1.3-2.2)	13.9
Antihypertensive medication	2.2 (1.7-2.7)	2.0 (1.6-2.5)	1.1 (0.8-1.5)	1.1 (0.8-1.5)	0.5
Obesity	4.6 (3.8-5.6)	4.5 (3.7-5.4)	2.6 (2.0-3.5)	2.6 (2.0-3.5)	44.3
Abdominal obesity	3.8 (3.2-4.5)	3.8 (3.2-4.6)	2.7 (2.1-3.5)	2.7 (2.1-3.5)	54.1
Cholesterol ≥6.5 mmol/L	1.4 (1.1-1.6)	1.2 (1.0-1.5)	1.1 (0.7-1.5)	1.1 (0.9-1.5)	0.8
Statin use	1.3 (0.9-2.0)	1.1(0.8-1.7)	1.1 (0.6-1.9)	1.1 (0.6-1.9)	0.1
Myocardial Infarction	1.5 (1.0-2.2)	1.4 (1.0-2.1)	1.4 (0.9-2.3)	1.4 (0.9-2.3)	2.0
Diabetes Mellitus	4.5 (3.1-6.5)	4.0 (2.7-5.9)	2.0 (1.2-3.3)	2.0 (1.2-3.2)	7.1
Family History	1.3 (1.0-1.6)	1.3 (1.0-1.5)	1.1 (0.9-1.4)	1.1 (0.9-1.4)	0.5
Smoking	1.1 (1.0-1.4)	1.2 (1.0-1.4)	1.5 (1.2-1.9)	1.5 (1.2-1.9)	14.5
Oral contraceptives	2.1 (1.8-2.6)	3.1 (2.5-3.8)	3.6 (2.7-4.7)	3.6 (2.7-4.6)	85.2
Horm.replacement therapy	1.1 (0.7-1.7)	0.9 (0.6-1.5)	1.3 (0.8-2.3)	1.3 (0.8-2.2)	1.0
Highest UAE quintile	1.9 (1.6-2.4)	1.9 (1.6-2.3)		1.1 (0.8-1.4)	0.3

Crude, age adjusted and multivariate gender-specific logistic regression models with and without elevated UAE in the model relating CV risk factors to the risk of having an elevated CRP level for that specific risk factor. The Wald statistic indicates the strength of the association.

BMI as well as waist circumference contributed most strongly to an elevated CRP in women as compared to men. This may be due to 1) a gender difference in the sensitivity of these measures of obesity to detect a surplus of overall and intra-abdominal fat or 2) a true gender-related difference in the relation between body fat and inflammatory cytokines. In the former case, it has been shown that for a given BMI, women have a higher percentage of total body fat relative to men.[31] Whether this holds true for waist circumference in relation to abdominal fat mass is uncertain. In the second case, the presence of an increased intra-abdominal fat mass in women may indicate a more pronounced state of insulin resistance relative to men, expressed as a concomitant rise of inflammatory proteins. Interestingly, recent data showed that females who were overweight or had a higher abdominal fat mass had an increased risk for hyperleptinemia as compared to men as well.[32]

Table 4. Multivariate logistic regression analysis: gender interactions.

Risk factors	B	S.E.	*P*
Age >50 yr	0.642	0.111	<0.001
Female	-0.166	0.159	0.296
SBP ≥140 and/or DBP ≥90	0.308	0.087	<0.001
Antihypertensive medication	0.187	0.107	0.080
Obesity	0.326	0.159	0.040
Abdominal obesity	0.368	0.139	0.008
Cholesterol ≥6.5 mmol/L	0.229	0.091	0.011
Statin use	-0.182	0.175	0.298
Myocardial infarction	0.380	0.137	0.006
Diabetes	0.776	0.159	<0.001
Family history	0.113	0.085	0.184
Smoking	0.858	0.104	<0.001
Oral contraceptives	1.265	0.137	<0.001
Hormone replacement therapy	0.243	0.277	0.379
UAE highest quintile	0.333	0.087	<0.001
Female * Age >50 y	-0.599	0.165	<0.001
Female * Obesity	0.640	0.215	0.003
Female * Abdominal obesity	0.601	0.192	0.002
Female * Smoking	-0.457	0.151	0.002
Constant	-2.654	0.109	<0.001

All variables are indexed as 0 and 1 (eg. male = 0 vs female=1, non-obese = 0 vs obesity=1 et cetera). B = β-coefficient, S.E.=standard error of B.

The finding that smoking is the most important determinant of an increased CRP in men is in agreement with data from Folsom *et al*, who showed that smoking was associated with an increased CRP in men but not in women.[11] However, conflicting data is coming from other studies showing CRP to be associated with smoking in women also, independent of other factors.[33]

The finding that oral contraceptive use, but not HRT, was related to an elevated CRP level is noteworthy. Most placebo-controlled studies clearly show an increase in CRP levels during HRT use over time.[34,35] This increase may be non-specific and unrelated to CV risk, since other markers of atherosclerosis show a decrease during HRT therapy.[35] Reasons for the absence of a relation between HRT and CRP in our study may be related to the cross-sectional design of the study and the low prevalence of HRT use in our female cohort.

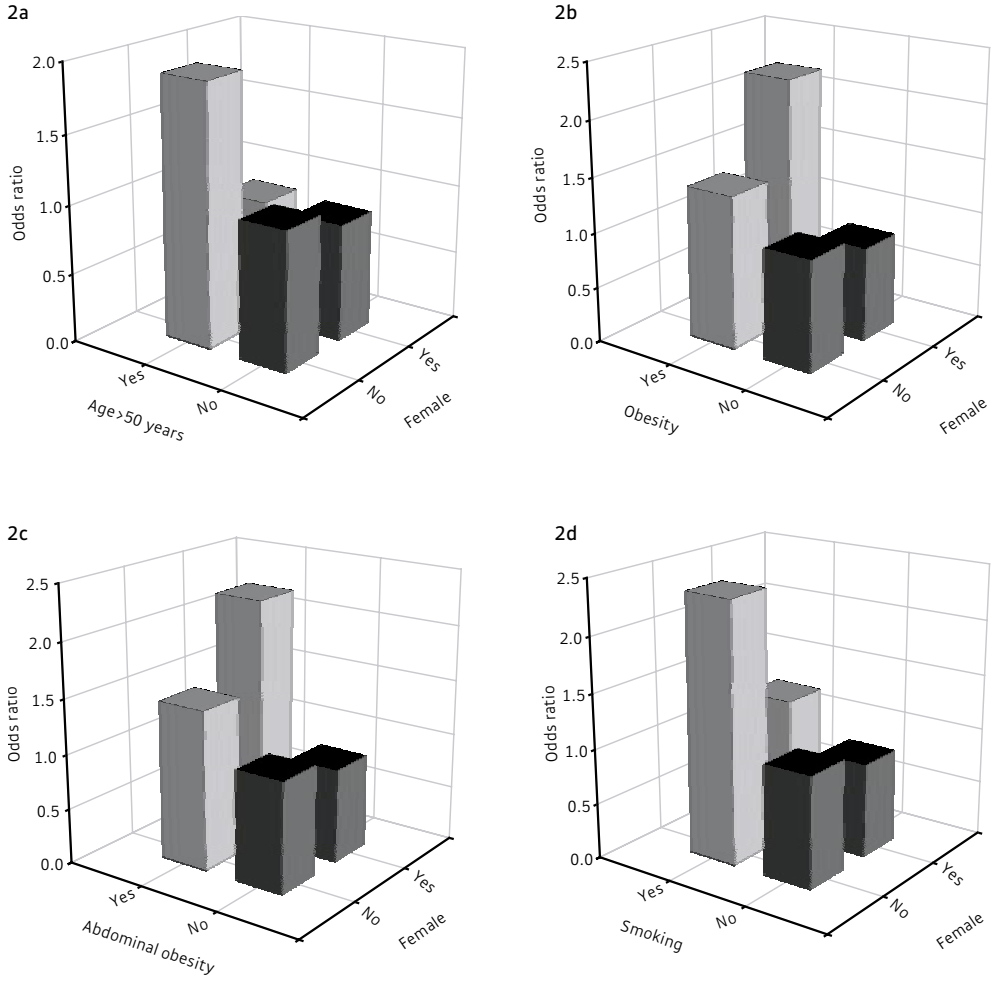
We, in addition, found that the associations between the known CV risk factors and CRP hardly changed when adding urinary albumin excretion in the model, neither in men nor in women. It has been shown that CRP and UAE independently predict CV mortality in (pre)-diabetic subjects,[36] which supports the concept of differentially involved pathways. This may be of clinical relevance because treatment can be focused on interventions that will result in a lowering of CRP levels or on a lowering of UAE. The present cross-sectional data raise the question whether correction of obesity and cessation of smoking will be the most effective method to correct elevated CRP,[37] whereas it is well known that antihypertensive treatment is the most effective method for reducing UAE.

The necessity of a specific gender dependent cut-off level to optimise CV risk assessment has been discussed in a number of recent reports.[25,26,38,39] Considering the slight difference in CRP levels between men and women, the use of a gender-specific cut-off point may be argued. Although CRP levels were quite similar, our results however show that age and risk factors clearly relate differently to CRP between the genders. This may have profound implications in CV risk assessment if CRP is included.

Of course, our study is only of a cross-sectional character. We found associations, but cannot conclude to a cause and effect phenomenon. In that respect we have to await the long-term follow-up of our subjects who are presently seen 4 years after the initial screening. We hope to confirm that during that follow-up period some CV risk factors will result more in a rise in CRP and that other risk factors will be associated with the development of an increased UAE, and that these changes over time are different for men and women. We also should realise that our cohort is not an aselect sample of the general population, but is enriched for the presence of an elevated UAE.[21] In our opinion, this however, does not distract from our conclusions. Our data were obtained from a large group of a wide age range, which makes the analyses robust. Moreover, it is a well-defined population with respect to the presence of CV risk factors.

In conclusion, CRP levels are higher in women than in men. The risk factors associated with an elevated CRP are different for men and women. In women, the use of oral contraceptives and measures of obesity were the strongest risk factors associated with a high CRP, while in men smoking is, besides age, the most important contributor to an elevated CRP. The presence of interaction terms with gender indicates that the impact of CV risk factors on CRP should be considered separately for both genders. Our data suggest that the difference in hormonal situation between men and women may explain higher CRP levels in women.

Figure 2a-d. Interactions: gender, risk factors and the risk of having an elevated C-reactive protein level.



a) Interaction between gender*age with the level of CRP

b) Interaction between gender*obesity with the level of CRP

c) Interaction between gender*abdominal obesity with the level of CRP

d) Interaction between gender*smoking with the level of CRP

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